

Plant Growth and Development

Panel Manager - Dr. Lon S. Kaufman, University of Illinois, Chicago

Program Director - Dr. Liang-Shiou Lin

This program area supports research aimed at increasing our basic understanding of mechanisms underlying the regulation of plant growth and development in order to achieve optimal productivity of agriculturally important crop and forest plants species. Research areas emphasized by the program include: (1) mechanisms of cell division, expansion and differentiation, (2) responses to environmental signals normally required for growth and development, (3) seed development and germination, (4) vegetative development, (5) reproductive development, (6) senescence and dormancy, (7) hormonal regulation of growth and development, (8) mechanisms of transducing internal and external signals required for normal growth and development, (9) cell biology, including studies on cytoskeleton, membrane transport, protein trafficking, and cell wall structure and properties.

2000-01657 Molecular Genetic Analysis of Potassium Nutrition in Plants

Zhu, J.

University of Arizona; Department of Plant Sciences; Tucson, AZ 85721

Grant 2001-35304-09892; \$130,000; 2 Years

Potassium nutrition is essential for plant growth and a key factor controlling crop productivity. Potassium uptake by plant roots is mediated through multiple transporters. The activities of potassium transporters are regulated by environmental factors such as potassium availability and sodium stress. The *Arabidopsis* protein kinase, SOS2, is a critical regulatory factor for potassium nutrition because mutations in the *SOS2* gene render the plant unable to grow on culture media with low levels of potassium or with high levels of sodium. The goals of this project are to characterize the *SOS2* gene and to identify proteins that interact with SOS2. The proposed studies will provide important insights into the molecular basis of the regulation of plant potassium nutrition.

2000-01438 Role of the *Arabidopsis* MONOPOLE Gene in Embryogenesis

Lukowitz, W.; Somerville, C.

Carnegie Institution of Washington; Department of Plant Biology; Stanford, CA 94305

Grant 00-35304-9394; \$163,000; 2 Years

The primary root of plants is formed early in embryonic development. Using *Arabidopsis* as a model organism for higher plants, we have isolated mutants that fail to establish a root at the correct position and time in embryogenesis. Despite this severe defect, some of the mutant embryos subsequently manage to form a fully functional root through what seems to be a regeneration-like process. Thus, it appears that the mutations specifically affect root initiation, i.e., the decision where and when to make a root. We have isolated the gene responsible for this defect in the mutants, designated *MONOPOLE*. The sequence of the *MONOPOLE* gene product is similar to known transcription factors, strongly suggesting a regulatory function for *MONOPOLE* in activating or repressing other genes. A detailed analysis of this gene may help us understand the molecular mechanisms organizing root initiation in the embryo as well as

in a more general context. This knowledge might facilitate manipulations of the root system in other, economically important species. The proposed research will focus on the following objectives: Determine at which time in development and in which cells *MONOPOLE* is active. Investigate possible links to other molecular signals implicated in root initiation, such as the plant hormone auxin. Investigate whether *MONOPOLE* is sufficient to trigger root formation. Investigate whether two closely related genes present in the genome serve overlapping or similar functions.

2000-01587 Molecular Analysis of *Arabidopsis* Floral Organ Abscission

Liljegren, S.J.

The Salk Institute for Biological Studies; San Diego, CA 92186-5800

Postdoctoral Fellowship; Grant 2001-35304-09897; \$90,000; 2 Years

Plants shed organs, such as leaves, flowers, and fruit, through the process of abscission. Abscission is a developmentally regulated process, with separation taking place within specialized cell types known as abscission zones. Surprisingly, although numerous studies have addressed the physiological aspects and hormonal regulation of the abscission process, very few studies have focused on the regulation of abscission zone development, and the genes required for abscission to occur. We plan to characterize an *Arabidopsis* mutant, *nevershed*, in which floral organ and leaf abscission is completely blocked. By using molecular markers expressed in the abscission zone at various stages of its development, we will be able to determine when abscission zone development is altered in the *nevershed* mutant compared to wild-type plants. We plan to identify the gene corresponding to the *nevershed* mutant, and to look for other genes which are involved in *Arabidopsis* abscission zone development. Advances made in understanding abscission in this model plant should be applicable to crops in which it would be desirable to control abscission. The ability to control abscission in fruit crops, and in important crops such as cotton and field beans, would allow increases in crop yield as well as simplify harvesting

2000-01539 Control of Endosperm Development by Imprinted Polycomb Genes

Fischer, R.L.

University of California, Berkeley; Department of Plant Biology; Berkeley, CA 94720

Grant 2001-35304-09891; \$130,000; 2 Years

The long term objective of this proposal is to understand the molecular mechanism that controls endosperm initiation and seed size. In flowering plants, the central cell of the female gametophyte is fertilized to produce the endosperm. Endosperm supports development of the dicot embryo, and in monocot plants, it comprises the majority of the seed mass and nutritional value. Genetic analysis indicates that a small number of imprinted activator and repressor loci have a profound effect on endosperm development and seed size. A model, termed the parental conflict theory, proposes that endosperm, and thereby seed size, requires the correct balance of activators and repressors. To test this model, we identified and cloned the MEDEA imprinted polycomb gene that is an endosperm repressor. To understand how endosperm and seed size are controlled, we will determine when the MEA gene is imprinted, how imprinting is maintained, and why imprinting affects only the endosperm, and not the embryo. We will identify DNA sequences that control MEDEA gene imprinting. Finally, we will

identify new imprinted genes that control endosperm development. These experiments will generate novel information about how polycomb proteins regulate reproduction, endosperm growth, and seed size.

2000-01457 ETTIN and the Gynoecium: Probing Auxin Signaling and Complex Morphogenesis

Zambryski, P.

University of California, Berkeley; Department of Plant and Microbial Biology;
Berkeley, CA

94720

Grant 2001-35304-09986; \$130,000; 2 Years

The gynoecium is the female reproductive organ of flowering plants, and is the most complex organ that flowering plants produce; however, the molecular mechanisms that coordinate gynoecium development currently are unknown. Our research focuses on one particular mutation, *ettin(ett)*, that provides a remarkable window for viewing many aspects of gynoecium development, as *ett* induced alterations affect differentiation in both longitudinal and transverse axes. Molecular cloning revealed that *ETT* encodes a transcription factor involved in auxin signaling. Thus, the function of the *ETT* gene links plant hormones to complex morphogenesis. The present studies aim to continue to dissect *Arabidopsis* gynoecium development using genetic, molecular, biochemical, and cell biological methods. We will assess the link between auxin signaling and morphogenesis, by testing the effects of chemically altering auxin signaling on gynoecium development (by applying auxin transport inhibitors), and performing genetic crosses between *ett* and known mutants in the auxin response pathway. Further, we will screen for suppressors and enhancers of the weak allele of *ett* to identify other genes in this pathway. Gynoecia develop into fruit, which account for approximately 91% of agricultural production for human consumption. Basic studies of gynoecium development are an important foundation for more applied research, in areas where transgenically improving fruit yield and composition, or altering compatibility and restricting fertility are desirable. Identifying, isolating, and characterizing genes involved in gynoecium development give a direct handle on engineering desirable traits into this structure.

2000-01434 Roles of Expansins and Cell Wall Hydrolases in Tomato Seed Germination

Bradford, K.J.

University of California, Davis; Department of Vegetable Crops; Davis, CA 95616-8631

Grant 2001-35304-09893; \$220,000; 3 Years

Seeds are the delivery system for improved crop genetics and for an expanding array of value-added traits developed through breeding and genetic engineering. As the value of seeds increases, methods to enhance seed performance or prevent undesired propagation are needed. Knowledge of the biochemical and molecular mechanisms underlying germination will lead to strategies for achieving these goals. In tomato seeds, weakening of the tissue enclosing the radicle tip (termed the endosperm cap) is required to allow the radicle to emerge from the seed and complete germination. The weakening process is associated with the production of enzymes that break down the cell walls of the endosperm tissue. In addition, a unique protein known as expansin is expressed

specifically in the endosperm cap prior to radicle emergence. Expansin proteins are involved in cell wall growth and in cell wall softening or separation processes. Expansins are thought to cooperate with cell wall enzymes to loosen the bonds that make the wall rigid, allowing it to expand or to be softened by the action of the enzymes. Using recombinant DNA techniques, we will develop transgenic tomato plants in which the expression of cell wall enzymes and expansins in the endosperm can be specifically promoted or inhibited. By determining the consequences of these changes on tissue weakening and germination, we will be able to determine which proteins are essential to the germination process. This will identify targets for either enhancing the germination of crop seeds or preventing the germination of weed seeds.

2000-01433 Function of the Ovule Regulatory Gene *INO*

Gasser, C.S.

University of California, Davis; Section of Molecular and Cellular Biology; Davis, CA 95616

Grant 2001-35304-09989; \$151,000; 2 Years

Ovules are the precursors to seeds - the major sources of protein and calories for both humans and livestock. Better understanding of ovule development can aid in producing more stable seeds with desirable storage and germination characteristics. Ovule development also serves as an attractive model to study plant development in general. We recently isolated the *Arabidopsis* *INNER NO OUTER (INO)* gene which is essential for formation of the outer ovule integument, a structure which goes on to form part of the seed coat. *INO* is a member a small gene family, the "YABBY" genes, which encode putative transcription factors. Other members of this family regulate polarity in leaves and floral organs. Chimeric proteins, made by swapping different regions between *INO* and other YABBY proteins, will be used to define the protein regions responsible for the different activities of the YABBY proteins. Models for the *in vivo* role of *INO* in ovule development will be tested through ectopic expression of *INO* under control of a variety of regulatory sequences which produce general or restricted expression patterns. Random DNA sequences will be screened to identify sites to which *INO* can bind. A library of *Arabidopsis* genomic DNA will also be screened to identify sequences from genes which are potential downstream targets regulated by *INO*. Together this work will help define the molecular mechanism of *INO* action, will identify new putative ovule regulatory genes, and will provide new inroads into the molecular basis of ovule, seed, and plant development.

2000-01586 Genetic Analysis of Rop GTPase Signaling in *Arabidopsis*

Yang, Z.

University of California, Riverside; Department of Botany and Plant Sciences; Riverside, CA 92521

Grant 2001-35304-09894; \$130,000; 2 Years

In plants, proper development and growth require coordinate communications between cells, tissues, and organs. Such communications involve cell surface receptors and subsequent intracellular signaling cascades. The goal of this proposed research is to understand how Rop GTPases, which belong to a class of guanine nucleotide binding proteins that act as important intracellular signaling switches, relay extracellular signals

from the putative cell surface receptor CLV1. CLV1 controls the maintenance of the shoot apical meristem (SAM) in *Arabidopsis*. Our preliminary studies have provided biochemical evidence that Rop may transmit signals CLV1. Two major objectives will be achieved in the proposed research. First, the specific Rop GTPase involved in the CLV1 signaling will be identified using combined functional genomics and genetic approaches. Second, we will use *clvl* mutant plants that are defective in CLV1 kinase activity to investigate whether CLV1 passes signals onto Rop through CLV-dependent phosphorylation of Rop, CLV1-dependent switching on Rop GTPase, and/or CLV1-dependent recruitment of Rop to the plasma membrane. The study described in this proposal will not only provide important basic understanding of signal transduction and molecular mechanisms for the control of meristem activity in plants, but also help to develop novel means to genetically manipulate plant signaling pathways for the improvement of crop production and food quality.

2000-01558 Molecular Bases of High-Affinity Potassium Transport and Na⁺ Transport in Plants

Schroeder, J.I.

University of California, San Diego; Division of Biology; Cell and Developmental Biology Section 0116; La Jolla, CA 92093-0116

Grant 2001-35304-09988; \$130,000.00, 2 Years

Potassium is one of the major macronutrients in higher plants required for plant growth and development. Potassium (K⁺) transport is important for many physiological processes including root and shoot growth, tropisms, cell expansion, enzyme activities, salinity stress and osmoregulation. Genes that encode putative molecular mechanisms for K⁺ transport in plants have been isolated. By expression cloning and functional analysis in heterologous systems several gene families have been identified, which may contribute to K⁺ uptake into plant cells. At least three gene families, have been proposed to contribute to K⁺ transport in plants including K⁺ channels, the wheat *HKT1* gene and the family of *AtKUP* (*ATKT* or *HAK*) transporters. The long term goal of this proposal is to gain insight into the physiological roles of cloned K⁺ transporter genes while focusing on a member of the *AtKUP-KT-HAK* family. Effects of mutation of the K⁺ transporter on plant growth and sensitivity to toxic cations will be analyzed. Furthermore, the expression pattern and cellular localization will be analyzed. Functional properties of the *Arabidopsis AtHKT1* gene will also be analyzed in plants by analyzing a disruption mutation, the expression pattern and the membrane localization of the transporter. Furthermore, the molecular basis for the unique sodium selectivity of *AtHKT1* will be analyzed. Results from these studies will lead to a molecular physiological understanding of potassium and sodium transport in plants and may contribute to development of future strategies for engineering improved K⁺ nutrition, growth and K⁺ uptake-related stress tolerance in crop plants.

2000-01554 The ABCD Model of Flower Organ Identify

Yanofsky, M.F.; Pelaz, S.

University of California, San Diego; Department of Biology; La Jolla, CA 92093-0116

Grant 2001-35304-09987; \$240,000; 3 Years; 2000 Award: \$74,000

Easily the most recognizable accomplishment in plant developmental biology is the proposal of the landmark ABC model of flower organ identity, where the individual

and combined activities of the ABC genes specify the identity of flower organs. We have characterized the closely related *AGL2*, *AGL4* and *AGL9* MADS-box genes from *Arabidopsis* which share overlapping expression patterns early in flower development. Our data indicate that *agl2 agl4 agl9* triple mutants display the striking phenotype in which all flower organs appear sepal-like. Thus, these three genes are functionally redundant and are required for petal, stamen and carpel development. The triple mutant appears indistinguishable from the previously described *bc* double mutants, suggesting that these genes are required for the activities of the previously described B and C genes. Based on these and other observations, we propose that the *AGL2/4/9* genes encode a D-function that is active in the three inner whorls where it specifies organ identity by interacting with B- and C-functions. In the current proposal, we plan to directly test our revised ABCD model of flower organ identity by establishing the role of the *AGL2/4/9* gene set in flower development. The specific objectives of this proposal are to (1) provide a detailed description of the *agl2agl4agl9* single, double and triple mutant phenotypes, (2) study the expression of organ identity genes in the triple mutant, (3) study the genetic interactions between *agl2/4/9* and the organ identity gene mutants, (4) determine if *AGL2/4/9* can interact with organ identity genes outside of the floral context, and (5) determine if the expression of the *AGL2/4/9* genes is controlled by the meristem identity genes.

2000-01441 Activation of *APETALA3* Floral Homeotic Gene Expression

Irish, V.F.

Yale University; Department of Molecular, Cellular and Developmental Biology; New Haven, CT 06520

Grant 2001-35304-09990; \$130,000; 2 Years

This project is aimed at understanding the molecular mechanisms responsible for flower development in *Arabidopsis*. While many genes involved in this process have been characterized, we still have very little understanding as to how key regulatory genes are appropriately activated in the floral meristem. This project will address how the floral homeotic gene *APETALA3* (*AP3*), which is required to specify petal and stamen identities, is regulated. *LEAFY*, a master regulator of flowering, appears to act in conjunction with another gene, *UFO*, to regulate *AP3* expression. We have preliminary evidence that *LFY* and *UFO* physically interact, and plan to carry out further biochemical and genetic experiments to test the relevance of this interaction for the regulation of *AP3*. In addition, preliminary screens have resulted in the identification of two other genes which may play critical roles in the activation of *AP3* expression. This project will also focus on analyzing the roles of these two new genes. Together, these studies should provide a more detailed understanding of the regulatory mechanisms involved in floral patterning. In addition, these studies have broader implications in that a better understanding of the regulation of floral development will be valuable for developing strategies to control the reproductive capabilities of agronomically important species.

2000-01655 Evolution and Expression of the *APETALA1* Gene Family in Angiosperms

Litt, A.

Yale University; Department of Molecular, Cellular and Developmental Biology; New haven, CT 06520

Postdoctoral Fellowship; Grant 2001-35304-09901; \$89,220; 2 Years

The ABC model of floral development postulates that the activity of specific combinations of genes in the four concentric domains of a developing flower specifies the identity of the organs that will form in each domain. The model is based on two model species, *Arabidopsis* and *Antirrhinum*. The gene *APETALA1* (*API*) of *Arabidopsis* is cited as an example of an A-group gene, required for sepal and petal identity. However, the actual role of *API* is somewhat open to interpretation, and there is little supporting evidence from other species, thus it is possible that A function as specified by the model does not exist. This project will investigate the evolution and the activity of members of the *API* gene family in an attempt to address questions about A-group function. Preliminary analyses suggest that there may have been duplication events in the *API* lineage that were important in the evolution of floral structure; this project will test this hypothesis via broad sampling of flowering plant species. Expression patterns will be investigated in a subset of species; these patterns can provide information related to gene function, thus it is important determine if they are conserved (and therefore if their function might be conserved). This information will help us assess the robustness and the limits of the ABC model. It is critical to know if the model is applicable beyond *Arabidopsis* and *Antirrhinum*, because it currently provides the framework for all research into floral structure and development in flowering plant species.

2000-01581 Characterization of VH1, a Provascular LRR Receptor-Like Kinase from *Arabidopsis*

Nelson, T.

Yale University; Department of Molecular, Cellular and Developmental Biology; New Haven, CT 06511

Grant 2001-35304-09992; \$130,000; 2 Years

Provascular cells give rise to the vascular tissues that form the venation of plant organs. In leaves, provascular cells arise from undifferentiated ground cells in a spatial pattern that later becomes the pattern of leaf veins. Provascular cells undergo cell divisions, elongation, and differentiation that are distinct from their neighboring cells. It is crucial to understand the origin and function of provascular cells because defects in the development of the vascular system have broad effects on plant physiology and development. However, little is understood of the means by which the network pattern of provascular cells is formed from a field of apparently equivalent ground cells, since few molecular markers or mutations have yet been identified that are specific for provascular cells. We identified the gene for a provascular-specific leucine-rich repeat (LRR) receptor-like kinase (RLK), by means of enhancer-trap screening in *Arabidopsis*. The gene, which we named *VASCULAR HIGHWAY1* (*VH1*), encodes a predicted protein with the domain structure typical of plant RLKs, including an extracellular ligand-binding domain and an intracellular serine/threonine kinase domain. On the basis of its similarity to other signaling kinases, its expression pattern, its antisense phenotypes, and its *in vitro* kinase activity, we propose that VH1 is a component of a provascular-specific signaling pathway that transduces intercellular information into downstream provascular

differentiation. We propose to characterize the role of VH1 in provascular cells by manipulating its expression in transgenic plants, and through a detailed study of the factors that influence its cell-specific expression.

2000-01542 The *Arabidopsis* GF14 / 14-3-3 Gene Family: Structure and Function

Ferl, R.J.; Sehnke, P.

University of Florida; Department of Horticultural Sciences; Gainesville, FL 32611-0690

Grant 00-35304-9601; \$270,000; 3 Years

The purpose of this work is to gain an understanding of the structure and function of the 14-3-3 proteins, which have emerged as major regulatory proteins in many aspects of cellular processes. Recent data from many species indicate that these proteins are involved in general regulatory control of cellular processes and signal transduction. Due to the presence of multiple 14-3-3s in plants, it is possible that these proteins have unique or specialized functions, and one emerging role is in the regulation of starch synthesis. With a thorough understanding of the structure and function of these proteins we should be able to understand several important biochemical pathways in plants, and possibly modify development for increased production or changes in the composition of secondary compounds without the need for increased culture, harvest or postharvest investments.

2000-01099 Anti-Microtubule Activity of a Carbamate Class of Plant Herbicides

Hugdahl, J.D.

Mercer University; Department of Chemistry; Macon, GA 31207

Seed Grant; Grant 00-35311-9596; \$46,913; 2 Years

Microtubules are polymers composed mainly of the protein tubulin. Tubulin, a dimeric protein composed of α - and β - subunits, associate to form hollow, filamentous microtubule polymers. These microtubules are dynamic structures, polymerizing and depolymerizing, as they form different functional arrays within the cell. Microtubules are required for vesicle transport, formation of spindles to separate chromosomes during cell division, and determination of cell polarity. In contrast to mammalian tubulins and microtubules, plant tubulins and microtubules are sensitive to small, simple, aromatic herbicides. Of the three major classes of plant herbicides, the carbamates appear to have a different mode of action, which may include non-tubulin targets. Understanding the interactions of herbicides with plant proteins will provide insights into how plant cells function. The purpose of this project is to fully understand the interactions of carbamate herbicides (specifically chloroisopropyl-*N*-phenyl carbamate, CIPC) with proteins in plant cells. The objectives of this project are to synthesize and characterize radiolabeled CIPC analogs, and characterize their interaction with plant tubulins and microtubules both *in vitro*, and in living cells. Photoaffinity analogs of CIPC will be used to localize herbicide binding sites by activating CIPC analogs in the presence of tubulin proteins using light. The activated CIPC analog will irreversibly bind to the protein. In a series of similar experiments the CIPC derivatives will be used to identify and characterize non-tubulin targets. Finding non-tubulin targets may allow identification of proteins that play an important role in cellular events such as cell division.

2000-01436 Coordinate Regulation of the Acetyl-CoA Carboxylase Genes During Seed Development

Nikolau, B.J.; Wurtele, E.S.

Iowa State University; Departments of Biochemistry, Biophysics and Molecular Biology and Botany

Grant 2001-35304-09991; \$130,000; 2 Years

The biosynthesis of fatty acids is a developmentally regulated metabolic process leading to the accumulation of a wide variety of products that directly or indirectly impact agriculture. In particular, a large portion of the fatty acids synthesized during seed development are deposited as seed oil, which is essential for the subsequent germination of the seed, and is also a major agricultural product. Acetyl-CoA carboxylase catalyzes the conversion of acetyl-CoA to malonyl-CoA, the first reaction in fatty acid biosynthesis. Considerable evidence, including our own research with *Arabidopsis*, indicates that plastidic acetyl-CoA carboxylase plays a critical role in controlling fatty acid biosynthesis and hence oil deposition. In *Arabidopsis*, plastidic acetyl-CoA carboxylase is a heteromeric enzyme, coded by four nuclear genes (*CAC1A*, *CAC1B*, *CAC2* and *CAC3*) and one plastidic gene (*accD*). We have isolated and sequenced all five of these genes and are characterizing the spatial and temporal pattern of their expression. Our data indicate that during seed development the spatial and temporal patterns of *CAC1A*, *CAC2*, *CAC3* and *accD* mRNA accumulation are coordinated and coupled with lipogenesis. We propose experiments that will test the hypothesis that this coordination of the accumulation of the *CAC* mRNAs is achieved by the coordinate regulation of the transcription of the three nuclear *CAC* genes. Ultimately this research will lead to a molecular understanding of the developmental regulation of seed oil biogenesis via the control of acetyl-CoA carboxylase gene expression. Such an understanding is essential for a rational molecular genetic improvement of oil seed crops.

2000-01204 Growth Chambers for Temperature Sensitive Mutants

Pickett, F.B.

Loyola University of Chicago; Department of Biology; Chicago, IL 60626

Equipment Grant; Grant 2001-35311-10189; \$24,170; 1 Year

Two temperature sensitive mutants, *arrested development 1* (*add1*) and *arrested development 3* (*add3*), of the plant *Arabidopsis thaliana* have dramatic effects on normal plant growth. Plants grown at high temperature (29°C) produce either no leaves (*add1*) or leaves missing internal cell types (*add3*). Precise control of growth temperature, made possible by the two Conviron Growth Chambers requested in this grant, will permit temperature up and downshift experiments that will reveal the developmental stage or stages during which each gene is required. Preliminary results from upshifts of *add1* plants indicate that the meristem, the source of all leaves, stems and flowers in mature plants, becomes enlarged and inactive following upshift. Questions we will ask of this mutant with our new chambers include determining the precise time that *add1* embryos first become sensitive to temperature induced meristem loss, and whether or not meristem function can be easily re-established following downshift. Similarly, initial experiments with upshifts performed on *add3* plants suggest that a wave of cell fate establishment, similar to that seen in the *Drosophila* eye, moves across the meristem during each round of new leaf production. Our new growth chambers will allow us to determine when in

leaf development this *add3* mediated response originates, and determine the approximate time of its duration. The chambers will also be used in new genetic screens to identify more mutants and will be used in functional genomic projects to discover genes requiring *add1* or *add3* expression for their normal transcription.

2000-01536 Characterization of Cell Wall Biogenesis Mutants of *Arabidopsis*

Carpita, N.C.

Purdue University; Department of Botany and Plant Pathology; West Lafayette, IN 47907
Grant 2001-35304-09993; \$130,000, 2 Years

We have developed Fourier transform infrared microspectroscopy as a high through-put method to identify mutations that specifically affect cell wall structure and architecture. By computer analysis of spectra we are able to trace certain types of mutations specifically to the cellulosic network or to the pectin gel matrix surrounding it. We have used this technique to amass from mutagenized populations of *Arabidopsis*, several mutants with alterations in cell-wall structure or architecture. A systematic protocol employing biochemical, spectroscopic, and imaging methods was developed to classify mutants into one of the six stages of wall synthesis and disassembly. We will apply this protocol to an established population of *Arabidopsis* cell wall mutants with deficiencies in pectin sugar components. We will also explore the impact of alteration of xyloglucan structure on biochemical and physical properties of the walls of three genetically defined mutants. A major practical goal is to generate plants with genetically defined variation in cell wall composition and architecture to permit assessment of these modifications on plant development. As cell walls are an enormously important source of raw material, we anticipate that several of the genes we identify and characterize, as well as several of the plants with genetically defined alterations, will be of economic importance. Examples include the modification of pectin-cross-linking or cell-cell adhesion to increase shelf life of fruits and vegetables, the improvement of yield and quality of fibers, and the relative allocation of carbon to wall biomass.

2000-01533 Characterizing Functions of Multiple Phospholipase Ds in *Arabidopsis*

Wang, X.

Kansas State University; Department of Biochemistry; Manhattan, KS 66506-3702
Grant 2001-35304-10087; \$ 130,000; 2 Years

Membrane phospholipid hydrolysis occurs in response to various cellular and environmental changes in plants. This activity may generate cellular messengers, change membrane composition, and/or degrade cellular membranes. Phospholipases mediate the first step in the hydrolysis and are key enzymes in the processes. The goal of this project is to understand the control and cellular functions of a major plant phospholipase family, phospholipase D (PLD). PLD has been proposed to be involved in multiple cellular functions, including seed germination, senescence, and stress responses, and the PLD family consists of several isoforms with different biochemical properties. To understand the function of the family members in plants, this project will determine: 1) cellular and tissue localization of different PLDs, 2) expression and intracellular movement of PLDs in response to environmental stress, and 3) the role of PLDs in plant response to water stress. Results of these studies will provide information about the involvement of

individual PLDs in specific cellular and stress conditions and also have potential to identify important steps for engineering plants with improved stress tolerance.

2000-01531 Analysis of Senescence-Specific Genes Using *Arabidopsis* Enhancer Trap Lines

Gan, S.

University of Kentucky; Department of Agronomy; Lexington, KY 40546-0091

Strengthening Award; Grant 2001-35304-09994; \$120,000; 2 Years

The last phase of plant leaf development is generally referred to as leaf senescence or aging. During senescence, chlorophyll and other leaf cellular components such as proteins are degraded, resulting in a sharp decline in the photosynthetic capability of the leaf. Therefore, leaf senescence limits crop yields and forest plant biomass production. Leaf senescence also contributes to postharvest loss of vegetable crops and devalues ornamental plants. Despite the importance of leaf senescence, the fundamental mechanisms underlying it are poorly understood. The long-term goal of the proposed research is to unveil regulatory mechanisms of leaf senescence, and based on the molecular findings, to genetically manipulate this process to achieve optimal and sustainable agricultural productivity. To approach this goal, we have identified more than one hundred *Arabidopsis* plants in which potential senescence-specific genes have been tagged by a molecular tag called T-DNA. We plan to first systematically analyze the regulation of all these potential senescence-specific genes by senescence-promoting factors, then to investigate the question whether these genes are expressed during senescence of other organs such as stems, flowers, and fruits in addition to leaves. We also plan to clone and characterize some of the senescence-specific genes to try to understand what they are, how they are regulated, and what roles they may play in senescence. This research will not only provide fundamental information on the molecular regulatory components and mechanisms of leaf senescence, but will also provide a means to devise genetic strategies to engineer senescence for agricultural application.

2000-01456 Seventh FASEB Summer Conference on "Mechanisms in Plant Development" Richards, E.J.; Irish, V.

Federation of American Societies for Experimental Biology (FASEB); Bethesda, MD

20814-3998 Grant 2001-35304-09929; \$5,000; 1 Year

The seventh FASEB Summer Research Conference on 'Mechanisms in Plant Development' will be held August 12-17, 2000 at the Vermont Academy in Saxtons River, Vermont. This conference will bring together biologists studying plant developmental mechanisms using genetic, genomic and cell biological approaches. In addition, we plan to highlight two emerging areas: the evolution of plant developmental programs, and the impact of epigenetic regulation on plant development and morphology. Beyond simply extending a successful conference series, we believe that this FASEB Summer Research Conference can serve two important roles for the scientific community. First, the meeting will provide an intimate venue conducive to creative interaction among researchers with different expertise and perspectives. This is particularly important as the maturation of molecular genetic research and the availability of genomic resources usher in a movement towards more integrated approaches

incorporating biochemical and cell biological data. Second, the FASEB conference will provide opportunity to attend a smaller meeting, where interaction between younger and more established researchers can occur. The conference will benefit the public by bringing together scientists from both industry and basic research to facilitate development of approaches for manipulation of plants toward agricultural goals.

2000-01432 Multiple Cellulases in *Arabidopsis* Flower Abscission

del Campillo, E.

University of Maryland, College Park; Department of Cell Biology and Molecular Genetics; College Park, MD

Grant 2001-35304-10088; \$130,000; 2 Years

Plants abscise organs by a process of cell-cell separation that takes place at the base of the organ to be shed. Many cell-wall hydrolases probably act in coordination to bring about abscission. Of the many types of hydrolases involved, cellulase (endo -1, 4 glucanase) genes are unique because of the contrasting role they can play depending on whether they are membrane associated or secreted. I hypothesize that membrane cellulase are likely to be involved in cell-wall synthesis and have to be down regulated prior to shedding. Consequently, suppressing their expression will accelerate abscission. This stage is followed by the up-regulation of secreted, abscission specific cellulases that contribute to the shedding and, consequently, the repression of their expression will delay abscission. The specific objective of the project is to determine the action of different cellulases during the process of abscission, using *Arabidopsis* as a model system. The long-term goal is understanding how plants alter their cell wall as part of many physiological processes. Abscission and other processes of cell separation are critical in determining the economic value of many important crops through effects on harvest, storage and marketability of agricultural products. Results from this research will increase our understanding of how cellulases work in plants and will help to decide whether, and how, this type of genes can be used to develop strategies to manipulate the timing of the abscission processes in a variety of plant systems.

2000-01569 The Role of bHLH Genes in Root Epidermal Cell Specification

Schiefelbein, J.W.

University of Michigan; Department of Biology; Ann Arbor, MI 48109-1048

Grant 2001-35304-10134; \$240,000; 3 Years; 2000 Award: \$74,000

Although poorly understood, the appropriate specification of cell identity is crucial for the growth and development of plants. The formation of the root epidermis of *Arabidopsis* provides an attractive model for the study of fundamental features of cell fate specification, because only two cell types are formed and they arise in a predictable spatial pattern. Indirect evidence has led to the notion that a bHLH-type transcription factor plays an important role in guiding epidermal cell fate in the *Arabidopsis* root. Recently, we and our collaborators have obtained direct evidence for key roles of three distinct bHLH proteins. These findings represent a breakthrough in our understanding of the molecular components involved in root epidermal cell specification, and we propose experiments to examine the role of each bHLH gene and their products. In brief, we plan to analyze their mutant phenotypes, their expression patterns, and their regulatory relationship to other known molecular components. This research is expected to lead to

new insights into the fundamental process of cell specification, and in particular, it will likely be viewed as a paradigm for spatial regulation of cell fate by multiple interacting transcription factors. Furthermore, a greater understanding of root epidermis development is expected to generate long-term improvements in agriculture by providing opportunities to manipulate root hair number to potentially enhance water and nutrient uptake by plants.

2000-01574 Genes Controlling Inflorescence Architecture in *Arabidopsis*

Sharrock, R.A.

Montana State University; Department of Plant Sciences; Bozeman, MT 59717

Strengthening Award; Grant 2001-35304-10084; \$120,000; 2 Years

The physical structure and stature of domesticated plants strongly influences their utility as food crops. Important agricultural properties such as pollination rates, the efficiency of harvesting, and overall plant productivity are dependent upon the structure of the inflorescence, the flower/fruit-bearing shoot. However, relatively little is known about the genetic control of the overall structure or "architecture" of the inflorescence. We are specifically interested in the molecular pathways that regulate the changes in growth and division of internode cells that are induced in many plants at the transition to the plant reproductive phase. These pathways control stem elongation in the inflorescence and, therefore, determine critical aspects of reproductive development. It may be possible to modify the activities of these pathways in targeted ways and, ultimately, to exert agriculturally-useful control over important aspects of plant structure. We have identified two genes that work together to regulate elongation of the inflorescence in the model plant *Arabidopsis*. These are called the *compact inflorescence* genes because mutations in these genes result in a striking lack of stem elongation in the flower shoots of the plant and formation of a cluster of flowers in place of the normally highly elongated floral bolt. The objectives of this project are to identify and clone these two genes and to develop a molecular understanding of how they function in controlling shoot elongation and growth.

2000-01453 Function of Auxin-Binding Protein 1

Jones, A.M.

University of North Carolina, Chapel Hill; Department of Biology; Chapel Hill, NC 27599-3280

Grant 2001-35304-10086; \$130,000; 2 Years

Auxin is a very important plant hormone because it is involved in many different aspects of plant growth and development such as seed production and germination, stem elongation, the number of shoots and roots, and the amount and size of fruit. An understanding of how this hormone regulates these different aspects of development is necessary for us to manipulate crop growth to yield more grain or fruit or to change the properties of plant growth to enable better agriculture. To address this, the receptor that recognizes auxin will be studied. This receptor is called auxin-binding protein 1 (ABP1) and previous results indicate that ABP1 mediates auxin-regulated cell expansion. Previous funding made it possible to identify a mutant plant that lacks this receptor and shown to be embryo lethal indicating its important role in early development. The mutant offers an opportunity to finally address the function of ABP1. In this project,

experiments will characterize the precise block in development in the mutant embryo at the morphological level (objective 1). This project will also create a plant in which the gene encoding for this receptor can be turned off at will (objective 2). An understanding of how this hormone works will lead to sustainable agriculture and is superior than application of chemicals that may be more costly and dangerous to humans and the environment. The original objectives are revised commensurate to reduced size of the award. The third objective to look for biochemical interactors with ABP1 has therefore been dropped.

2000-01540 The Regulation of Ethylene Biosynthesis in *Arabidopsis*

Kieber, J.J.

University of North Carolina, Chapel Hill; Department of Biology; Chapel Hill, NC 27599

Grant 2001-35304-10083; \$130,000; 2 Years

The gaseous hormone ethylene has profound and agriculturally significant effects on plant growth and development. Here we propose to continue our studies of the mechanisms regulating ethylene biosynthesis in etiolated *Arabidopsis* seedlings. Employing a simple seedling response to ethylene as a facile genetic screen, we identified a number of mutants that are affected in the regulation of ethylene biosynthesis. Our studies of these mutants have led to the hypothesis that post-translational modifications of ACC synthase, which catalyzes the rate-limiting step of ethylene biosynthesis, play an important role in regulating ethylene production. Furthermore, we have found that cytokinin regulates ethylene biosynthesis through one member of the ACS gene family, ACS5, in *Arabidopsis*. The studies supported by these funds will determine the nature of the post-translational modification of the ACS5 protein in both our mutants and in response to cytokinin, which may reveal a novel mechanism of control of this biosynthetic pathway. Furthermore, the *ETO3* gene will be cloned. Mutations in *ETO3* lead to large increases in the level of ethylene biosynthesis, and thus the sequence of this gene may provide clues to how ETO3 modulates the function of the ACC synthase enzyme, and thus the level of ethylene production. These studies should provide insight into the post-translational mechanisms regulating ethylene biosynthesis, which may ultimately lead to an enhanced ability to control the production of this key plant hormone in an agricultural setting.

2000-01095 Isolation of Cellulose Synthase Genes from Economically Important Conifers

Klein, A.S.

University of New Hampshire; Department of Biochemistry and Molecular Biology; Durham, NH 03824

Research Career Enhancement Award; Grant 2001-35311-10120; \$78,892, 1 Year

Wood fiber consists primarily of two macromolecular components, lignin, a random polymer of phenolic compounds, and cellulose, a crystalline polysaccharide of repeating beta-1,4-linked glucose residues. When wood is processed to produce paper, sulfuric acid is used to remove lignin. These paper pulping processes pollute waterways. Conifers are important sources of paper pulp. Several laboratories are investigating how conifers produce lignin. Among their goals are to reduce the amount of lignin produced

in normal wood formation. The other half of the biological equation would be to increase the proportion of wood fiber that consists of cellulose. Recently the structural genes for cellulose have been identified in cotton, *Arabidopsis*, and corn. The amino acid sequences of these proteins are highly conserved making it possible to identify corresponding cellulose synthase genes in conifers. The overall goal of this project is to isolate conifer cellulose synthase genes, so we can begin to characterize regulation of cellulose biosynthesis in conifers in comparison to the regulation of lignin synthesis. The specific objectives are 1) to design conserved oligonucleotide primers to amplify fragments of the gene with the polymerase chain reaction 2) Use these primers amplify specific full length cDNAs from white pine and radiata pine messenger RNA libraries; these are putative pine cellulose synthase clones. 3) Use biochemical assays to verify cDNAs are actually homologous to cellulose synthase genes from cotton and *Arabidopsis*. 4) Use these clones to isolate full length genes (regions coding for protein and regulatory regions).

2000-01455 Engineering Pathogen Resistance by Manipulating Salicylic Acid Biosynthesis and Metabolism

Raskin, I.

Rutgers, the State University of New Jersey; Cook College; Center for Agriculture and the Environment; New Brunswick, NJ 08901

Grant 2001-35304-10085; \$130,000; 2 Years

Salicylic acid is (SA) an important signal that regulates disease resistance in plants. A systemic increase in SA levels elicited by pathogens induces resistance to a broad spectrum of diseases. This resistance is associated with the accumulation of pathogenesis-related (PR) proteins that are also induced by SA. As a result of previous USDA funding we have advanced the understanding of SA signaling, elucidated the pathways of SA biosynthesis and metabolism in tobacco, and characterized and cloned the key enzyme in SA metabolism UDP-glucose:salicylic acid 3-*O*-glucosyltransferase (SA-GTase). Successful cloning of SA-GTase has given us the tools to engineer plants with increased SA levels and resistance to various diseases. This will be accomplished by blocking the expression of SA-GTase. Engineered plants will be tested for their resistance to agronomically important pathogens. Ability to manipulate SA levels in plants will also allow us to ask fundamental questions about the role of SA in plant growth and development. We will also characterize and clone SA-GTase promoter, and assess its suitability as an effective and fast-acting inducible promoter useful for the regulation of agronomically important transgenes. The ability to regulate recombinant gene expression in crop plants with simple, cost effective and environmentally safe stimuli, such as SA, is one of the important targets of plant biotechnology. The proposed research may lead to the development of a novel, environmentally friendly and cost-effective strategy for increasing plant resistance to pathogens while improving our knowledge of biological processes regulated by SA.

2000-01572 *Ramosa1* Regulates Stem Cell Fate in the Malze Inflorescence

Martienssen, R.; Vollbrecht, E.

Cold Spring Harbor Laboratory; Cold Spring Harbor, NY 11724

Grant 2001-35304-10133; \$240,000; 3 Years; 2000 Award: \$72,000

The familiar ear of maize, known as the female inflorescence, is an unbranched even-rowed structure. In other cereal crops such as millet or sorghum, the inflorescence is branched giving rise to many more seed. In 1912, a mutant of maize was discovered that had a multiply branched ear and tassel, and was named "*ramosa*". It was originally thought to be a new species of corn, but was subsequently shown to be the result of a mutation in a single gene. We have now molecularly isolated the DNA sequence that codes for the *ramosa* gene, and we are studying its role in branching and seed set. The gene encodes a likely transcription factor that regulates other genes and so we are pursuing genetic and genomic approaches to identifying these "targets". We will characterize the plant cells in which the gene functions in normal and mutant corn. *Ramosa* is highly conserved in other plants and we will explore whether variation in the *ramosa* gene is responsible for diversity of plant architecture among the grasses. We hope to be able to exploit these discoveries in manipulating this key trait leading to higher yields in many different crops.

2000-01450 Nuclear Import of Nucleic Acid-Protein Complexes in Plants

Citovsky, V.H.

State University of New York; Department of Biochemistry and Cell Biology; Stony Brook, NY 11794

Grant 00-35304-9333; \$236,000; 3 Years

Traffic of nucleic acid-protein complexes into the cell nucleus is a basic process which, nevertheless, is still not well understood in plants. We study nuclear import of nucleic acid-protein complexes using plant infection by *Agrobacterium tumefaciens*, a soil pathogen known to genetically alter plants by transferring a segment of its DNA (T-DNA) into the host cell nucleus. This study will help to understand and control the infection process and increase nuclear uptake of the DNA of interest in genetic engineering experiments. Also, since pathogens, such as *Agrobacterium*, often adapt existing cellular machinery for their own needs, our work will contribute to the understanding of plant nuclear import in general. The proposed research will seek to identify and characterize plant proteins and their encoding genes that recognize *Agrobacterium* T-DNA-protein complexes (T-complexes) and mediate their nuclear import. Specifically, we shall elucidate the molecular pathways used for T-complex nuclear import that share similarity with animal and yeast nuclear import as well as those that are specific for plants and do not exist in non-plant systems. Identification of plant genes required for the T-complex nuclear import will result in new technologies for genetic manipulation of agronomically important plants that are presently recalcitrant to *Agrobacterium*-mediated transformation. On the other hand, inactivation of the same genes in susceptible plants will make them resistant to *Agrobacterium* infection.

2000-01926 Intercellular Protein Trafficking and Leaf Development

Ding, B.

Ohio State University; Department of Plant Biology; Columbus, OH 43210

Grant 2001-35304-09928; \$130,000; 2 Years

The phloem tissue serves as the pipeline to transport photoassimilates and other types of molecules throughout a whole plant. Accumulating evidence suggests that the phloem transports proteins and RNAs which may play a role in regulating various plant

functions such as cell growth, organ formation, flowering, and interactions with pathogens. How a specific protein or RNA is taken up by the phloem for long-distance transport and is then dispatched to target cells to perform its functions is virtually unknown. Using the movement protein of cucumber mosaic virus as a tool to characterize intercellular and phloem transport of proteins, we have obtained data to support the hypothesis that complex mechanisms operate to regulate protein transport between the phloem and surrounding nonvascular tissues. In this project, we seek to test this hypothesis by: 1) identifying the specific cellular boundary that controls protein trafficking between the phloem and surrounding nonvascular tissues, and 2) developing a genetic system to identify the genes involved in the regulation. Results from this project will provide initial insights about the mechanisms underlying regulated protein transport across the phloem-nonvascular interface and establish a foundation for further studies to elucidate such mechanisms at the molecular level. Knowledge of this nature is essential to understand how plant growth and developmental processes and plant-pathogen interactions are regulated at the whole plant level. It may also be valuable in designing strategies to genetically modify crops to obtain desirable traits such as controlled carbon allocation, flowering and resistance to pathogens.

2000-01566 G-Protein Function in Guard Cells: A Genetic/Cell Biological Approach

Assmann, S.M.

The Pennsylvania State University; Department of Biology; University Park, PA, 16802
Grant 2001-35304-09916; \$220,000; 3 Years

Heterotrimeric G-proteins, composed of α , β , and γ subunits, are signal-transducing proteins that play a central role in such vital processes as vision, taste, and olfaction. Approximately 50% of the drugs used in clinical medicine target cellular pathways containing G-protein elements. Plants also possess G-protein subunits, but their function in plant systems is poorly understood. Two plant G-like genes, *GPA1* and *AtXLG1*, are expressed in guard cells. Guard cells are specialized cells located in the outermost cell layer of the plant. Swelling and shrinking of pairs of guard cells regulate the size of microscopic pores ("stomata") in the plant surface through which both carbon dioxide uptake and water loss occur. Thus, guard cells control both photosynthetic rate and plant water status. In the proposed research, the effects on guard cell function of genetic manipulation of *GPA1* and *AtXLG1* protein levels will be assessed using several physiological assays. In particular, the role that these G-proteins play in translating plant water stress into a guard cell response that closes the stomata (thus reducing water loss under drought conditions) will be assessed. It will also be determined where within the plant cell *AtXLG1* is located. Animal G-proteins both bind and hydrolyze (break down) the substrate, GTP, in a cycling mechanism. *AtXLG1* protein will be assayed to determine whether it also exhibits these biochemical characteristics. A greater understanding of the function and biochemical nature of these plant G-proteins will contribute to efforts to improve plant drought tolerance.

2000-01579 ABA Signal Transduction by Phospholipase D

Gilroy, S.

The Pennsylvania State University, Department of Biology; University Park, PA 16802
Grant 2001-35304-09898; \$130,000; 2 Years

The development and germination of seeds is controlled by plant hormones. In barley, the hormone gibberellin promotes germination whereas abscisic acid inhibits this process. Failure of correct hormonal regulation of germination can lead to major economic crop losses through, for example, premature germination or preharvest sprouting. The broad goal of this research is to characterize the molecules that translate the abscisic acid signal to the cellular activities in the seed that prevent germination. We have previously characterized that in barley, abscisic acid activates the enzyme phospholipase-D that then triggers the abscisic acid response pathway in the seed. Abscisic acid does not directly activate this enzyme, implying a series of molecules lie between an abscisic acid receptor and phospholipase-D. This proposal therefore seeks to determine how abscisic acid regulates the phospholipase-D through functional analysis of the structure of this enzyme. A second goal of the research is to define the nature of the signal transduction elements that lie between the abscisic acid receptor and phospholipase-D. This analysis will concentrate on the role of G-proteins, a ubiquitous class of signaling intermediates known to act in other hormone regulated pathways in plants and animals. By understanding how such specific molecules are involved in the abscisic acid-regulated system that modulates seed germination, it should be possible to develop strategies to manipulate grain quality or alleviate major seed-associated crop losses such as pre-harvest sprouting in cereals.

2000-01583 Gordon Conference on Plant Senescence, Abscission, and Programmed Cell Death

Bleecker, A.B.

Gordon Research Conference; Gordon Research Center; West Kingston, RI 02892-0984
Grant 96-35304-5317; \$5,000; 1 Year

Senescence and other forms of programmed cell death are extremely important processes in the development and life history of plants. Yet our understanding of the molecular mechanisms that drive these processes lags behind that for the developmental processes. The Gordon Research Conference on Plant Senescence has been held every four years and has highlighted developments in this field since its inception many years ago. The last Conference expanded the coverage to include programmed cell death in the programs. For the 2000 Conference, we intend to continue this tradition with an emphasis on the impact of progress in the areas of genetics, genomics and molecular biology. The following 8 topic areas will be covered: Developmental Programmed Cell Death; Stress-Induced Programmed Cell Death; Cell Biology, Biochemistry, and Physiology of Senescence; Molecular Biology and Genetics of Senescence; Abscission and Dehiscence; Hormonal Control Systems; Meristem Fate; and Genomic Approaches and other Hot Topics.

2000-01443 Gordon Research Conference on Plant and Fungal Cytoskeleton

Cande, Z.W.

Gordon Research Conferences; Gordon Research Center-University of Rhode Island, West Kingston, RI 02892-0984

Grant 2001-35304-09913; \$5,000; 1 Year

A Gordon Research Conference on the Plant and Fungal Cytoskeleton will be held August 13-18, 2000 in Andover, New Hampshire. The cytoskeleton is intricately

involved in mitosis, cytokinesis, polarity determination, organelle placement, and directed cell expansion and motility. Many advances in understanding of cytoskeleton function resulted from studies in plants and fungi and identified promising avenues for treatment of cancer, control of fungal pathogens, and for development of strategies to increase crop productivity. The conference brings together active researchers to communicate recent results, engage in discussion, and plan collaborative projects. The conference organizers widely surveyed the research community to identify the most exciting work being done currently. Emphasis in the program is placed on biological processes such as mitosis and cytokinesis, morphogenesis, cell communication and polarity determination, and related signal transduction processes. In addition, speakers will present research that makes use of innovative techniques in real-time analysis of dynamic cellular processes, genetics and molecular manipulation. The program includes speakers from Japan, the United Kingdom, Australia, Germany and the United States. Whenever possible, junior level investigators were selected to make presentations. Much of the external funding obtained to support the conference will be used to defray expenses of students and postdocs.

2000-01452 Gordon Research Conference on Plant Cell Walls

Delmer, D.P.

Gordon Research Conferences; Gordon Research Center-University of Rhode Island;
West Kingston, RI 02892-0984

Grant 2001-35304-09914; \$3,000; 1 Year

In August 2000, a Gordon Conference on Plant Cell Walls will be held at Kimball Union Academy in Meriden, New Hampshire. Cell walls play a key role in plant growth and development, are a key resource for the timber and textile industries, and also represent rich sources of polymers for food products, chemicals, paper and packaging. Cell walls also play important roles in disease resistance and prevention of water loss and as a major competing sink for reduced carbon. The conference is both timely and important since the past few years have witnessed an explosion of new knowledge about walls as the power of molecular biology has taken hold for workers in this field. Genes involved in the synthesis of major wall polymers are being rapidly identified and characterized in model plants, agricultural crops and in trees. Having such genes in hand and understanding their patterns of expression and the consequences of modifying levels of expression, allows one to manipulate cell wall compositions in ways we could not have dreamed just a few years ago. This meeting brings together international scientists from academia, government and industrial labs who work on wall structure, identification of genes involved in wall synthesis, assembly and growth, topology and crystal structures of enzymes encoded by these genes, selection and characterization of mutants impaired in wall synthesis, and experts in genomics. The relatively isolated and intimate site for this Gordon Conference is ideal for such a conference that aims to stimulate interactions between scientists with these diverse approaches to the topic.

2000-01142 Equipment Request to Strengthen Basic Infrastructure for Plant Biology Research at the University of Rhode Island

Chandlee, J.M.; Kausch, A. P.

University of Rhode Island; Department of Plant Sciences; Kingston, RI 02881

Equipment Grant; Grant 96-35304-5318; \$24,610; 1 Year

Over the last two years there has been a growing emphasis on transgenic turfgrass research within the internationally recognized turfgrass program at the University of Rhode Island involving faculty collaborations between molecular biologists, breeders, pathologists and others. In addition, curricular revision in the Department of Plant Sciences at the undergraduate level has produced a revised undergraduate major in "Environmental Plant Biology" designed to provide a strong foundation in plant molecular biology and biotechnology. At the heart of this new curriculum is a learner-centered pedagogy that encourages extensive involvement of undergraduates in faculty research programs. In support of this concept, a recently awarded USDA Higher Education Challenge Grant supports undergraduate interns who will participate in faculty research projects centered on plant transgenics during the summer months. These activities have placed new demands on the existing plant growth facilities at URI. This request initiates our 3-year plan to establish a fully functional, comprehensive Plant Growth Facility at URI. Funds from this current equipment grant request will help to initiate the process of acquiring the necessary growth chamber equipment for this work and alleviate the current high demand for existing growth chamber space by students and faculty. We intend to purchase a Conviron plant growth chamber (Model PGR15) which will be used to support research activities utilizing plant gene transfer technology at URI. This improvement to the research infrastructure will make users more competitive for extramural funding opportunities in the future on projects that use the new biotechnologies for genetic enhancement of plants.

2000-01530 A Regulator of Lateral Root Formation in *Arabidopsis thaliana*

Bartel, B.

Rice University; Department of Biochemistry and Cell Biology; Houston, TX 77005

Grant 2001-35304-09925; \$220,000; 3 Years

Auxins are plant hormones that promote lateral and adventitious root formation, and auxins are widely used in agriculture for this purpose. However, our understanding of the primary targets of auxin action in the plant remains incomplete. The discoveries that the auxin-regulated *Aux/IAA* genes encode short-lived nuclear proteins that can interact with a family of transcription factors provided potential identities of the direct regulators of auxin responses. Analyzing the roles of individual *Aux/IAA* family members, which probably perform both overlapping and unique functions during development, is essential to understanding auxin responses in plants. The subject of this proposal is *IAA28*, a newly-discovered member of the *Aux/IAA* gene family. Increasing the activity of this gene leads to profound changes in plant development that are suggestive of auxin response defects. Specifically, it appears that one of the normal function of *IAA28* is to prevent or delay lateral root formation. The proposed experiments will elucidate the role of *IAA28* in plant development. We will analyze the expression of the *IAA28* gene in wild-type and mutant plants both throughout development and in response to alterations in hormone levels. We will determine the localization and stability of the *IAA28* protein and mutant versions of the protein. Finally, we will examine the phenotypic consequences of decreasing *IAA28* protein levels. In addition to contributing to our basic knowledge of the factors that control developmental and environmental auxin responses,

a detailed understanding of these factors may provide insights for their modification in agriculturally important plants.

2000-01459 Molecular Dissection of the Vernalization Pathway

Amasino, R.

University of Wisconsin, Madison; Department of Biochemistry; Madison, WI 53706

Grant 2001-35304-10089; \$220,000 3 Years

Plants ensure that flowering occurs at a favorable time of year by sensing a number of environmental cues such as daylength and temperature. In many plant species flowering is promoted by an extended period of cold (i.e., winter) allowing plants to flower in favorable conditions of spring. This promotion is known as vernalization. Currently, we understand very little of how vernalization acts to promote flowering. In our research, we are using genetic studies with the model plant *Arabidopsis thaliana* to identify the genes involved in vernalization. In addition to increasing our understanding of how plants sense and respond to their environment, manipulating the requirement for vernalization could lead to significant improvements in crop plants. Sugar beets, for example, require cold treatment for flowering to occur. Typically, when planted in the spring sugar beets grow vegetatively all season developing a swollen tap root. If crops are planted too early in the spring, however, or if the spring is unusually cold, the sugar beets may become vernalized and go on to flower. When flowering occurs, sugars and nutrients stored in the tap root are mobilized to the flowers and developing seeds, significantly reducing the value of the crop. By creating sugar beets that are less sensitive to vernalization it may be possible to eliminate losses due to flowering and extend the growing season by allowing crops to be planted earlier in the spring.

2000-01235 Regulation of Ethylene Signal Transduction in Carnation Petals

Verlinden, S.

West Virginia University; Division of Plant and Soil Sciences; Morgantown, WV 26506-6108

Seed Grant; Grant 2001-35311-09996; \$54,584, 2 Years

The phytohormone ethylene plays an important role in the growth and development of plants. One of the many developmental processes in which ethylene plays a key role is flower senescence. In many plants flower senescence is associated with a large increase in ethylene production. This climacteric ethylene has been shown to play a regulatory role in the events leading to the demise of the flower. In addition to the mandatory perception of ethylene to complete the senescence program large increases in ethylene responsiveness have been observed during petal development. The ultimate goal of this lab is to understand the processes associated with the initiation of flower senescence. Identification and manipulation, by means of genetic engineering and/or applied chemicals, of some key elements in this process could lead to improved productivity (increased seed set) and uses (flower longevity) of agriculturally important crops. Little is known, however, about ethylene signal transduction in petals, a process central to understanding the initiation and maintenance of ethylene production, and how it mediates differences and changes in ethylene responsiveness. Research on ethylene signal transduction in *Arabidopsis* has made great strides in our understanding of ethylene perception and signaling in plants. One observation in particular, a

transcriptional cascade in ethylene signaling, has the potential to explain several of the experimental observations of flower senescence. We hypothesize that a transcriptional cascade in ethylene signaling operates in petals and is the key element in the increasing ethylene production that results in the ethylene climacteric and flower senescence.

2000-01439 Genetic Analysis of Cell Patterning during Maize Leaf Development

Sylvester, A.W.

University of Wyoming; Department of Botany; Laramie, WY 82071-3165

Strengthening Award; Grant 2001-35304-09899; \$120,000; 2 Years

Proper cell division is essential for the normal growth and development of all organisms. When cell division goes awry, tumors and malignancies characteristic of cancer can result. Plants have the remarkable ability, however, to adjust for abnormal cell division so that genetic defects are not as severe in plants as in animals. We are interested in understanding how this occurs by studying the genes that regulate cell division in corn plants. Using genetics as well as cellular and molecular methods, we have learned that groups of cells can adjust for abnormal cell division by expanding more and thereby maintaining normal cell shape and function. In addition, we have identified an important signaling protein that is involved in regulating cell division in plant cells. When this protein is absent, mutant plants have small tumors, similar to warts on the leaf, but have no other developmental problems. We have also found that many genes are involved in coordinating growth of neighboring cells and our research will help to discover the function of the proteins encoded by these genes. If we can identify specific proteins that regulate cell growth in plants, we will be able to design strategies to engineer or breed for improved growth or for specific traits in agronomically important crops.